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Original Article

Evaluation of Different Amounts of Pelleted Manure with Urea and Micro Elements on Yield and Seed Quality of Medicinal Plant, Pumpkin (*Cucurbita Pepo* Var.Styriaca)

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ABSTRACT

Objective: In this study investigated effects of different amounts of pelleted manure with urea and micro elements on yield and seed quality of medicinal plants, pumpkin (*Cucurbita pepo* var. styriaca). **Methods:** The animal manure and urea as pellet with four levels (150 Kg urea, 50 Kg urea + 3.5 ton animal manure, 100 (Kg) urea+ 1.5 ton animal manure and 150 (Kg) urea+1.5 ton animal manure) located in main plots and microelements with three levels (1000, 2000 and 3000 ppm) including a combination of iron, zinc, manganese and boron were located in subplots. Results showed that the highest number of female flowers, fruit yield, number of seeds per fruit, 1000 seed weight and seed yield was observed in using of 150 (Kg) urea + 1.5 ton manure pellet Increasing of nitrogen levels increased plant growth parameters, yield components and seed yield in pumpkin. **Results:** There was no significant difference between pelleted manures in seed oil percentage and seed protein but the highest oil yield was obtained in the treatment of 150 (kg) urea + 1.5 ton manure pellet which is related to higher seed yield in this treatment. The highest amounts of mentioned parameters are obtained in concentration of 2000 ppm of microelements except of seed protein which is observed in a concentration of 3000 ppm. At whole among of these fertilizer treatments, 1.5 ton animal manure + 150 (Kg) Urea as pellet with 2000 ppm of microelements is much recommended for maximum qualitative and quantitative properties in pumpkin.

1.INTRODUCTION

Manure produced from dairy cattle is by-product of livestock production system. The manure with high moisture and low density is not suitable to use. The densification of dry manure is the best method for decreasing the volume of manure which also decreases the costs of handling and storage (Alemi et al, 2010). Biomass densification means to use some form of mechanical pressure to reduce the volume of grind material and conversion of this material to a solid form, which is easier to handle and store than original material

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(Mani et al, 2006). Pelleting and cubing have been used for animal feeds and biomass wastes (Hernandez et al, 2006). Nitrate can be lost by denitrification and leaching, particularly under wet soil conditions. In addition both ammonium and nitrate can be tied up through immobilization by microorganisms in the soil as they decompose low-N organic residues (Grant, 2004; Rad et al, 2013). Increasing use of nitrogen fertilizer in agricultural production has raised concerns, because the N surplus is at risk of leaving plant-soil system causing environmental contamination and also increased costs associated with the manufacture and distribution of N fertilizer (Alizadeh and Ghadeai, 2006). So using of slow-release fertilizers decrease the leaching of N from the rhizosphere and will help to sustainability of agricultural systems. Ahmed et al (2007) reported that slow-release of nitrogen fertilizer increase the yield of *Sorghum bicolor*. Rymar et al (1989) reported that slow release fertilizers (N as encapsulated urea) are more effective that the conventional fertilizers. The split application of N fertilizers is cheaper, but slow-release N fertilizers, when applied at the right time, minimize the risk of N loses by leaching (Ahmed et al, 2007).

Currently, pumpkin seed oil is not widely used commercially even though it has characteristics that are well suited for industrial applications and can contribute to healthy human diets (Stevenson, 2003).The highly unsaturated fatty acid composition of pumpkin seed oil makes it well-suited for improving nutritional benefits from foods. Pumpkin seed oil has been implicated in providing many health benefits (Fu et al, 2006). The most critical health benefit attributed to pumpkin seed oil is preventing the growth and reducing the size of the prostate (Tasi et al, 2006). There is also evidence that suggests pumpkin seed oil can retard the progression of hypertension (Zuhair et al, 2000) and mitigate hypercholesterolemia (Zuhair et al, 1997) and arthritis (Fahim et al, 1995). Medicinal pumpkin (*Cucurbita pepo* var. styriaca) is an annual plant belonging to cucurbitaceae family. Pumpkin has been used as a vegetable and medicine since ancient times, but has been cultivated as a medicinal plant only in recent decades. Nowadays it is cultivated all over the world for different kind of usage. The fatty oil of seeds is also used as a local specialty, e.g., in Austria and Germany as salad dressing additive (Sigmund and Mukovic, 2004). The seeds of pumpkins contains fatty oil, β-sitosterol and E-vitamin and is used as a raw material for certain pharmaceutical products including Peponen and Gronfing capsules which are mainly used to cure the prosatic hypertrophy and urinary tract irritation (Stevenson et al, 2007). A combination of several ingredients and particular content of sterols are generally considered to contribute to the pharmacological activity (Sabudark, 2007).

The objective of this research was to study the effects of different amounts of Pelleted manure with urea and micro elements on growth, seed yield and oil quality of medicinal pumpkin.

2. MATERIALS AND METHODS

To study the effects of urea and manure as pellet and some microelements on seed yield and seed oil of medical pumpkin (*Cucurbita pepo* var. styriaca), a split plot experiment based on a randomized complete block design with three replicates was conducted at the research farm of Abouraihan agricultural college of Tehran University (Latitude:33°,28/N, longtitude:51°,46/ E).The animal manure and urea as pellet with four levels (150 Kg urea, 50 kg urea+3.5 ton animal manure, 100 Kg urea+1.5 ton animal manure and 150 Kg urea+1.5 ton animal manure) located in main plots and microelements with three levels (1000, 2000 and 3000 ppm) including a combination of iron, zinc, manganese and boron equally were located in subplots. To investigate the physico-chemical properties of the farm, samples are taken from 0-30cm and 30-60cm depths of the soil of the research plots. The cow manure used in this study was obtained from the animal husbandry from the college around the Tehran. The dry manure (7.6%w.b) and urea was ground using a hammer mill before pelleting. The mixture of manure with urea was pelleted by a pelleting machine (extruder type). After pelleting, the mixed pellets were studied for quality parameters like durability and constant of the nutrient content (Alemi et al, 2010). Seeds are cultivated at 21 May 2010 in a 1.5*0.4 m distances. Pelleted manure is used based on the treatments before planting. Seeds are germinated 7 days after planting. Plants were irrigated every 5 days until flowering and after that with 7 days irrigation interval. Weeds were controlled by hand.

2.1. Morphological, growth and yield parameters

For measurement of growth and morphological parameters, four plants were selected randomly in each treatment and parameters like number of branches, number of female flowers (in 50% flowering stage), plant length and chlorophyll content using SPAD502 (every 20 days) were recorded. Manual harvesting was carried out 110 days after planting when 75% of fruits became yellowish-orange in color and seeds were dark green and well rounded. yield parameters like seed yield, number of seeds per fruit, fruit yield, not ripened seed percentage, 1000 seeds weight, seeds weight per fruit, number of fruits per plant, fruit weight and width of fruits were recorded.

2.2. Oil extraction

The oil content of the seeds was determined by treating the weighted powder seeds with hexane and refluxed for 16 h in a Soxhlet extractor. The solvent was removed by rotary evaporator at 60 ºC. The oil sample was then placed in a vacuum oven kept at 60 ºC for 30 min and then accurately weighted and the percentage yield calculated (Younis et al, 2000).

2.3. Fatty acid composition

The fatty acid composition of oil extracted from pumpkin seeds was analyzed by injecting fatty acid methyl esters into a Shimadzu, Model: 14-A gas chromatogram equipped with a flame ionization detector and an RT-2560 column (100m×0.25 mm i.d., 0.20μm film thickness); temperature programming, 100-240 ºC at 3ºC/min; injector temperature 225ºC; detector temperature, 250 ºC; carrier gas, N_2 at rate of 1.1 ml/min.

2.4. Protein content

Content of crude protein in seeds was determined by Kjeldal method the method consists of three steps: 1) digestion of the sample in sulphuric acid with a catalyst. The nitrogen contained in the sample is converted to ammonia; ammonium sulphate being formed. 2) distillation of ammonia released from ammonium sulphate by addition of an excess of sodium hydroxide; ammonia being trapped in a trapping solution (sulphuric acid). 3) back-titration of the excess of the trapping solution (Naumann and Bassler, 1976).

2.5. Micro elements contents

Fe and Zn contents in seeds and shoot were determined using flame ionization atomic absorption, Spectrometer (model 1100 B of Perkin Elmer) as according to the method of Chapman and Pratt (Gomez and Gomez, 1984).

2.6. Statistical analysis

The data were exposed to the statistical analysis of variance by one-way ANOVA using Statistical Analysis System (2001) (SAS) and mean were compared by Duncan's multiple range test at 5% probability level.

3. RESULTS AND DISCUSSION

The physico-chemical properties of the soil and chemical properties of the cow manure are shown in table1 and table 2. Our results showed that different amounts of pelleted manure with urea significantly affected plant growth parameters, yield and yield components in medical pumpkin (Table3). The highest plant length and number of branches was obtained in the treatment of 150kgurea+1.5 tons manure pellet. A higher plant length and also more branches in a runner plant like pumpkin leads to higher leaf area index, more light absorption, and consequently higher photosynthesis and concentration of carbohydrates. The highest number of female flowers, fruit yield, number of seeds per fruit, 1000 seed weight and seed yield was observed in using of 150kgurea+1.5 tons manure pellet (Table5). Increasing of nitrogen levels increased plant growth parameters, yield components and seed yield in pumpkin. Gholipoori et al, (2007) and stepleton et al, (2000) reported that nitrogen levels had a significant effect on fruit numbers, mean weight of fresh fruit, 1000 seeds weight and seed yield in *Cucurbita pepo*. Nitrogen fertilizers had a great role in enhancing the metabolism processing due to the importance of nitrogen in building carbohydrates, protein and fats in plant tissues. Results (Table 5) indicated that in higher nitrogen levels the growth parameters, yield components and seed yield are increased. The higher seed yield in higher amount of nitrogen fertilizer attributed to higher number of seeds per fruit. The higher yields were probably responsible for better development of fruit, increased uptake of nutrients in plants leading to enhanced chlorophyll content and carbohydrate synthesis, higher accumulation of photosynthates and their distribution to developing ovules. These results are in conformity with findings of Ali et al (1999) in pumpkin.

Some studies reported that the appearance of first fruits in Cucurbitaceae plants prevent formation of new fruits (Robinson, 1993; Mercels, 1992). The first fruit in plant is a strong sink and compete with other plant parts for limited carbohydrates and in consequence prevent the formation of new fruits. Nitrogen in urea form can be lost rapidly by leaching, particularly under wet soil conditions. Therefore plant can be faced to nitrogen deficit in this developing phase which plant strongly needs to nutrients for formation of new fruits. Using of slow-release fertilizers decreases the leaching of N from the rhizosphere and plant can reach suitable amount of nitrogen, relevant to the developing phase need. Better plant growth, higher number of fruits per plant and seeds per fruit and in consequence higher seed yield in treatment of 150kg urea+1.5 ton manure-pelleted in compare to 150 kg/ha urea can prove that pelleting of manure with urea can be a suitable method to prevent nitrate leaching. The positive effect of pelleting in prevent of nitrate leaching can also be observed in SPAD data. The data (Table 5) showed that Chlorophyll content in 20 days after planting is higher in 150 $kg.ha^{-1}$ urea in compare with other treatments but at 60 days after planting it was no significant difference in chlorophyll content between treatments and at 80 days after planting, the chlorophyll content was higher in treatment of 150 kg urea + 1.5 tons manure pellet which can confirm the slow release of nitrogen in pelleted fertilizer. Mahdavi et al, (2004) and Takebe et al, (1990) reported that a positive correlation between Chlorophyll content and nitrogen available content in the soil. Using of slow release nitrogen fertilizers especially in pelleted forms with manure as an organic source for nitrogen delayed the ripening of fruits in pumpkin which is reported also by Gholipoori et al, (2007). There was no significant difference between pelleted manures in seed oil percentage and seed protein but the highest oil yield was obtained in the treatment of 150 kg urea + 1.5 ton manure pellet which is related to higher seed yield in this treatment (Table5). Jariene et al, (2007) reported that different amount of nitrogen fertilizers had no significant effect on seed oil percentage of Pumpkin. The same results are reported by Jahan et al, (2006). They observed that different amount of manure did not affected oil percentage in pumpkin.

Micro elements significantly affected some of growth parameters, yield components, yield and quality parameters in pumpkin like number of female flowers per plant, number of seeds per fruit, 1000 seeds weight, fruit yield, seed yield ,seed oil content , oil yield and seed protein content (Tables 3 and 4). The highest amounts of mentioned parameters are obtained in concentration of 2000 ppm of microelements except of seed protein which is observed in a concentration of 3000 ppm (Table 5). The positive effects of foliar spray of micro elements on yield, yield components and seed oil content in sun flower was reported by Thalooth et al, (2005). We observed that use of 2000ppm of microelements can hold the chlorophyll content in a higher amount at 80 days after planting in compare with other treatments which means higher photosynthesis and higher concentration of carbohydrates (Table 5).

Proteins are easily absorbed by humans (in rate of 65-80%) and appear to be the main building material for cells of human body. Seeds of oil-bearing pumpkins are a good source of crude proteins and can accumulate 35-40% of these compounds (Tarek et al, 2001). Our results showed that the content of seed protein in pumpkin can be increased significantly by using of 3000ppm of micro elements (Table 5). It is established that use of micro elements increased the content of crude proteins in seeds (Jariene et al, 2007).

Results showed that pelleted manure with urea treatments significantly affected the content of nutrients of zinc and iron in plant and seeds of pumpkin (Table 4). The highest amount of these two nutrients is obtained in treatment of 150 kg urea + 1.5 ton manure pellet (Table 6). Previous data indicated that pumpkin plants which received urea gained the best quality of fruit and the highest value of nutritional elements (N, P, K, Fe, Zn and vitamin C) (Jariene et al, 2007).The reported amount of zinc in pumpkin seeds was 1.09 mg in 100 g dried seeds (Mohamad, 2004). Our results showed that with use of pelleted manure with urea or microelements the zinc and iron contents and in consequence the nutritional value of pumpkin seeds can be enhanced.

The interaction effect of different amounts of pelleted manure with urea and micro elements affected significantly percentage of not ripened seeds and zinc density in medical pumpkin (Table 7). The lowest amount of not ripened seed is observed in treatment of 150 (Kg) urea + 1.5 tons manure (pellet) + 3000 (ppm) micro fertilizers. The highest density of zinc in plant biomass was observed in the same treatments. The interaction effect was not significant on other parameters. Analysis of several Styrian pumpkin seed oil revealed that the content of polyunsaturated fatty acids (45.6±5% rel) is considerably higher than the content of monounsaturated fatty acids (35.9±10% rel) or saturated fatty acids (18.5±20% rel) (Fruhwirth et al, 2003), with linoleic acid, oleic acid, palmitic acid, and stearitic acid (together≥98%) being the predominant fatty acids. The high content of linoleic acid is an important nutritional aspect of Styrian pumpkin seed oil since linoleic acid is an essential n-6 fatty acid. The result of interaction effect of different amounts of pelleted manure with urea and micro elements on the concentration of the predominant unsaturated fatty acids showed in Fig 1. Our results showed that the relative amounts for linoleic acid, oleic acid, palmitic acid, and stearitic acid are 55.89%, 31.47%, 10.66% and 1.41%, respectively (Fig. 1). The highest amount of linoleic acid was obtained in the treatment of 100 kg urea + 1.5 tons manure pellet + 2000 ppm micro elements. This improvement can be related to better nutrition condition in consumption of this fertilizer that it is due to gradual release of nitrogen and good effect of animal manure, although with considering of low consumption of animal manure as pellet and results of two other treatments, the role of gradual release of nitrogen from fertilizer has most important effect. At whole among of these fertilizer treatments, 1.5 tone animal manure + 150 (Kg) Urea as pellet with 2000 ppm of microelements is much recommended for maximum qualitative and quantitative properties in pumpkin.

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Table1.
Physico-chemical properties of the soil

Depth (cm)	Sand (%)	Silt (%)	Clay (%)	pH	EC ($ds.m^{-1}$)	P (ppm)	K (ppm)	OC (%)	Total N (%)	Fe (ppm)	Zn (ppm)	Mn (ppm)	Cu (ppm)	B (ppm)
30	56.4	21.4	23.2	7.7	4.15	45.8	288	0.75	0.44	3.9	1.07	3.88	0.78	0.6
60	79.8	9.1	12.6	7.6	4.09	40.2	216	0.15	0.42	3.1	0.87	3.06	0.71	0.54

Table2.
Physico-chemical properties of the manure

Total N (%)	Total P (%)	Total K (%)	OC (%)	pH	EC ($ds.m^{-1}$)	Cu ($mg.kg^{-1}$)	Zn ($mg.kg^{-1}$)	Fe ($mg.kg^{-1}$)	Mn ($mg.kg^{-1}$)
1.97	0.64	2.04	25.3	8.74	18.21	20.4	86.6	5612	180.4

Table 3.

Analysis of variance of the effect of different amounts of pelleted manure with urea and micro elements on growth parameters, yield and yield components of medical pumpkin

S.O.V	df	MS												
		Fruit yield	Seed yield	Oil yield	No. of seed per fruit	Not ripened seed percentage	Seed dry Weight per fruit	1000 seeds weight	No. of fruit per plant	No. of fruit per m^2	Weight of each fruit	Width of each fruit	Length of plant	No. of branches
R	2	35810320^{ns}	877.75^{ns}	199.61^{ns}	654.53*	11.31**	19.65^{ns}	0.14^{ns}	0.09^{ns}	0.14^{ns}	28779.88^{ns}	0.82^{ns}	243.81^{ns}	2.53^{ns}
A	3	661790787**	317152.27**	74717.50**	13036.85**	21.58**	452.32**	454.02**	0.21*	0.34*	646248.69**	3.58^{ns}	9694.1**	103.87**
Ea	6	17095276	1672.16	1257.72	167.32	1.70	3.81	16.49	0.01	0.02	11843.26	0.11	190.72	2.15
B	2	86021842*	36664.75**	22919.55**	1925.4**	49.74**	20.34^{ns}	83.34**	0.03^{ns}	0.05^{ns}	37628.72^{ns}	0.85^{ns}	108.58^{ns}	0.71^{ns}
A×B	6	10013023^{ns}	4273.62^{ns}	1385.26^{ns}	64.01^{ns}	3.58**	3.63^{ns}	5.02^{ns}	0.01^{ns}	0.02^{ns}	4466.02^{ns}	0.60^{ns}	31.65^{ns}	0.23^{ns}
R×B	4		3822.11				8.04		0.04	0.02	18614.31	0.37	19.17	1.41
Eb	16	14017778		900.49	18.138	0.56		8.79						
CV%		5.3	7.8	7.8	5	2.8	11.3	1.6	11.4	11.4	9.9	6.2	6.9	20.8
)A(														
(A1)	2					8.46**								
(A2)	2					24.66**								
(A3)	2					3.87*								
(A4)	2					23.48**								
CV%	2					3.6								

Table 4.

Analysis of variance of the effect of different amounts of pelleted manure with urea and micro elements on number of female flowers, Chlorophyll content and seed quality parameters of medical pumpkin

S.O.V	df	MS											
		No. of female flower	Spad (20 days)	Spad (40 days)	Spad (60 days)	Spad (80 days)	Spad (100 days)	Seed protein percentage	Seed oil percentage	Iron density in biomass	Iron density in seed	zinc density in biomass	Zinc density in seed
R	2	0.32^{ns}	0.004^{ns}	0.68^{ns}	0.63^{ns}	1.46^{ns}	6.5^{*}	1.67^{ns}	2.49^{ns}	260.90^{**}	4.93^{**}	41.16^{**}	0.71^{ns}
A	3	3.83^{**}	9.48^{*}	15.97^{**}	3.33^{ns}	26.51^{**}	34.62^{**}	0.21^{ns}	16.76^{**}	256.21^{**}	14.07^{**}	124.89^{**}	5.38^{**}
Ea	6	0.07	0.39	0.08	1.44	1.45	0.85	2.58	8.2	19.37	1.23	3.39	0.88
B	2	2.77^{**}	1.11^{ns}	0.19^{ns}	0.03^{ns}	14.9^{*}	16.0^{**}	38.97^{**}	58.22^{**}	44.48^{**}	7.80^{**}	48.71^{**}	1.82^{*}
A ×B	6	0.32^{ns}	0.26^{ns}	0.86^{ns}	1.19^{ns}	0.81^{ns}	3.35^{ns}	0.98^{ns}	0.81^{ns}	3.56^{ns}	0.37^{ns}	5.09^{*}	0.21^{ns}
R ×B	4	0.07	2.33	2.55		1.39	0.24				4.3		
Eb	16				1.27			0.75	1.222	1.93		1.91^{ns}	0.33
CV %		8.3	3.1	7.8	2.7	4.6	5.1	2.4	2.4	4.7	3.3	5.3	13.5
)A (													
(A 1)	2											6.66^{*}	
(A 2)	2											14.1^{**}	
(A 3)	2											3.2^{ns}	
(A 4)	2											40.02^{**}	
CV	2											6.2	

%	

^{NS}, nonsignificant or significant at $^{*}P \le 0.05$, and $^{**}P \le 0.01$.

Table 5.
Effect of different amounts of pelleted manure with urea and micro elements on growth parameters, yield, yield components and oil production in medical pumpkin

Treatments	Mean ±SE										
	Width of each fruit (cm)	Length of plant (cm)	No. of branches	No. of female flower	Spad (20 days)	Spad (40 days)	Spad (60 days)	Spad (80 days)	Spad (100 days)	Seed protein (%)	Seed oil (%)
Pelleted manure and urea											
150 (kg) Urea	20.9±0.3^{a}	292.1±4.2^{b}	10.4±0.4^{b}	4.7±0.1^{b}	43.1±0.3^{a}	45.3±0.3^{a}	45±0.3^{a}	36.8±0.6^{b}	25±0.5^{c}	35.5±0.8^{a}	45.1±0.6^{a}
50 (kg) Urea + 3/5 tons Manure (pellet)	19.5±0.1^{a}	249.8±5.2^{c}	7±0.3^{c}	3.6±0.1^{c}	40.6±0.3^{c}	42.4±0.1^{c}	45.6±0.2^{a}	36.1±0.5^{b}	23±0.3^{d}	35.6±0.8^{a}	42.1±0.5^{a}
100 (kg) Urea + 1/5 tons Manure (pellet)	20.1±0.2^{a}	283.3±3.2^{b}	9.3±0.4^{b}	4.7±0.1^{b}	42.0±0.3^{b}	42.7±0.3^{c}	41.3±0.4^{a}	38.6±0.6^{a}	26.4±0.7^{b}	35.7±0.5^{a}	44.7±0.9^{a}
150 (kg) Urea + 1/5 tons Manure (pellet)	20.7±0.4^{a}	329.6±5.9^{a}	15.1±0.8^{a}	5.2±0.2^{a}	42.3±0.3^{a}	44.1±0.3^{b}	42.4±0.3^{a}	39.9±0.4^{a}	27.5±0.5^{a}	35.3±0.6^{a}	44.7±1^{a}
Micro fertilizer											
1000 ppm	20.3±0.2^{a}	291±9^{a}	10±0.9^{a}	4.4±0.2^{b}	42.3±0.3^{a}	43.6±0.5^{a}	41.6±0.4^{a}	37.2±0.5^{b}	24.4±0.6^{b}	33.9±0.5^{c}	43.8±0.5^{b}
2000 ppm	20.6±0.3^{a}	289.9±10.5^{a}	10.6±1^{a}	5.1±0.2^{a}	41.7±0.3^{a}	43.8±0.3^{a}	41.6±0.2^{a}	39.1±0.5^{a}	26.7±0.7^{a}	35.3±0.3^{b}	46.5±0.8^{a}
3000 ppm	20±0.2^{a}	285.3±8^{a}	10.2±1^{a}	4.2±0.1^{b}	42.0±0.4^{a}	43.6±0.4^{a}	41.5±0.3^{a}	37.2±0.6^{b}	25.3±0.4^{b}	37.5±0.4^{a}	42.1±0.5^{c}

Treatments	Mean ±SE								
	Oil yield (Kg.ha^{-1})	Seed yield (Kg.ha^{-1})	Fruit yield (Kg.ha^{-1})	No. of seeds per fruit	Seed dry Weight per fruit (gr)	1000 seeds weight (gr)	No. of fruit per plant	No. of fruit per m^2	weight of each fruit (gr)
Pelleted manure and urea									
150 (kg) Urea	392.27±16.5^{b}	867.02±26.7^{b}	66082±1613.8^{c}	255.44±5.3^{a}	38.50±1^{a}	181.86±1^{b}	1.7±0.02^{b}	2.25±0.02^{b}	2921.1±52.6^{a}
50 (kg) Urea + 3/5 tons Manure (pellet)	260.61±11.1^{c}	616±20.3^{c}	61316±1347^{c}	182.50±5.1^{c}	24.12±1^{c}	176.8±1.2^{c}	2.05±0.09^{a}	2.57±0.09^{a}	2402.3±79^{b}
100 (kg) Urea + 1/5 tons Manure (pellet)	392.1±14.9^{b}	873.87±21^{b}	73985±1096.6^{b}	231.76±4.9^{b}	34.38±0.7^{b}	184.08±1.4^{b}	2.03±0.06^{a}	2.55±0.06^{a}	2907.0±61.1^{a}
150 (kg) Urea + 1/5 tons Manure (pellet)	481.82±21^{a}	1075.05±30.4^{a}	80727±2012.8^{a}	269.0±6.4^{a}	39.74±1.4^{a}	193.75±1.2^{a}	2.16±0.06^{a}	2.71±0.06^{a}	2976.4±65.8^{a}
Micro fertilizer									
1000 ppm	363.02±26.6^{b}	824.4±58.4^{b}	69594±2854.6^{b}	220.81±10.6^{b}	33.53±2^{a}	182.52±3.4^{b}	2.4±0.06^{a}	2.4±0.06^{a}	2828±79^{a}
2000 ppm	431.64±27.8^{a}	921.93±51.4^{a}	73547±2415.4^{a}	245.62±11.2^{a}	35.68±1.9^{a}	187.16±3.4^{a}	2.5±0.05^{a}	2.5±0.05^{a}	2839.8±87^{a}
3000 ppm	350.44±20.3^{b}	828.08±44.1^{b}	68442±2128.7^{b}	237.66±9.6^{a}	33.34±1.1^{a}	182.68±3.4^{b}	2.5±0.07^{a}	2.55±0.07^{a}	2737.4±97^{a}

Values with different letters denote differences at the 5% level of significance in the duncam test for each comparison between treatments in the respective column.

Table 6.
The effect of different amounts of pelleted manure with urea and micro elements on concentration of zinc and iron in shoot and seed of medical pumpkin

Treatments	Mean ±SE			
	Iron density in biomass (mg/100gr)	Iron density in seed (mg/100gr)	zinc density in biomass(mg/100gr)	zinc density in seed (mg/100gr)
	Pelleted manure and urea			
150 (kg) Urea	30.92±1.5^{ab}	14.31±0.4^{a}	22.81±0.9^{b}	4.83±0.2^{a}
50 (kg) Urea +3/5 tons Manure(pellet)	23.56±0.9^{c}	12.47±0.4^{b}	18.35±0.8^{c}	3.27±0.1^{b}
100 (kg) Urea +1/5 tons Manure(pellet)	26.86±1.9^{bc}	14.79±0.3^{a}	20.34±0.7^{c}	4.11±0.2^{ab}
150 (kg) Urea +1/5 tons Manure(pellet)	35.93±1.9^{a}	15.36±0.4^{a}	27±1.1^{a}	4.96±0.3^{a}
	Micro fertilizer			
1000 ppm	27.14±1.8^{b}	13.5±0.3^{c}	19.94±0.9^{c}	3.84±0.2^{b}
2000 ppm	30.78±1.9^{a}	14.09±0.5^{b}	22.51±1.2^{b}	4.46±0.2^{a}
3000 ppm	30.78±2^{a}	15.10±0.3^{a}	23.91±1.2^{a}	4.57±0.2^{a}

Values with different letters denote differences at the 5% level of significance in the duncam test for each comparison between treatments in the respective column

Table 7.
The interaction effect of different amounts of pelleted manure with urea and micro elements on percentage of not ripened seed and zinc density in medical pumpkin

Treatments	Mean±SE	
	Not ripened seed (%)	zinc density in biomass(mg/100gr)
150 (kg) Urea+1000 (ppm) Micro fertilizer	26.76±0.6 cd	21.20±1.9 cde
150 (kg) Urea+2000 (ppm) Micro fertilizer	25.46±1.5 de	23.07±3.6 bc
150 (kg) Urea+3000 (ppm) Micro fertilizer	23.43±0.7 g	24.15±2.4 b
50 (kg) Urea+3/5 tons Manure(pellet)+1000 (ppm) Micro fertilizer	30.4±0.8 a	16.49±2.3 g
50 (kg) Urea+3/5 tons Manure(pellet)+2000 (ppm) Micro fertilizer	29.16±0.1 ab	17.84±1.4 gf
50 (kg) Urea +3/5 tons Manure(pellet)+2000 (ppm) Micro fertilizer	24.93±0.4 ef	20.73±2.6 de
100 (kg) Urea +1/5 tons Manure(pellet)+1000 (ppm) Micro fertilizer	26±1 de	19.16±3.1 ef
100 (kg) Urea +1/5 tons Manure(pellet)+2000 (ppm) Micro fertilizer	25.7±0.9 de	21.06±1.7 cde
100 (kg) Urea +1/5 tons Manure(pellet)+3000 (ppm) Micro fertilizer	23.9±0.6 fg	20.8±2.1 de
150 (kg) Urea +1/5 tons Manure(pellet)+1000 (ppm) Micro fertilizer	28±0.2 bc	22.92±1.6 bcd
150 (kg) Urea +1/5 tons Manure(pellet)+2000 (ppm) Micro fertilizer	23.86±0.6 fg	28.1±1.2 a
150 (kg) Urea +1/5 tons Manure(pellet)+3000 (ppm) Micro fertilizer	22.66±0.3 g	29.98±1.5 a

Values with different letters denote differences at the 5% level of significance in the duncam test for each comparison between treatments in the respective column.

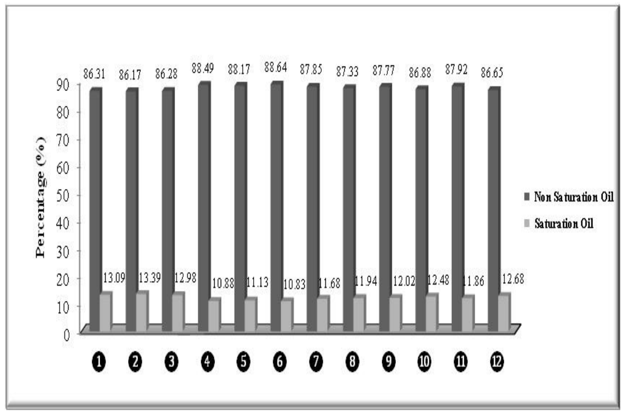
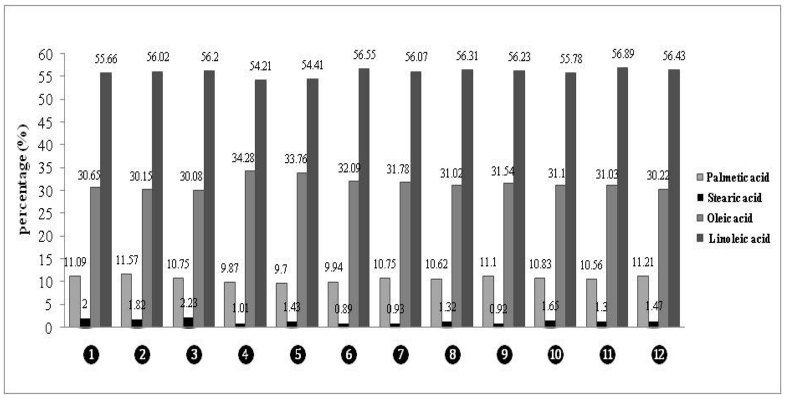


Fig 1. Interaction effect of different amounts of pelleted manure with urea and micro elements on the quality of seed oil in medical pumpkin



❶ 150 (kg) Urea+1000 (ppm) Micro fertilizer
❷ 150 (kg) Urea+2000 (ppm) Micro fertilizer
❸ 150 (kg) Urea+3000 (ppm) Micro fertilizer
❹ 50 (kg) Urea+3/5 tons Manure(pellet)+1000 (ppm) Micro fertilizer
❺ 50 (kg) Urea+3/5 tons Manure(pellet)+2000 (ppm) Micro fertilizer
❻ 50 (kg) Urea +3/5 tons Manure(pellet)+2000 (ppm) Micro fertilizer
❼ 100 (kg) Urea +1/5 tons Manure(pellet)+1000 (ppm) Micro fertilizer
❽ 100 (kg) Urea +1/5 tons Manure(pellet)+2000 (ppm) Micro fertilizer
❾ 100 (kg) Urea +1/5 tons Manure(pellet)+3000 (ppm) Micro fertilizer
❿ 150 (kg) Urea +1/5 tons Manure(pellet)+1000 (ppm) Micro fertilizer
⓫ 150 (kg) Urea +1/5 tons Manure(pellet)+2000 (ppm) Micro fertilizer
⓬ 150 (kg) Urea +1/5 tons Manure(pellet)+3000 (ppm) Micro fertilizer